be influenced by several genetic factors.[6] The higher rate in the males might be accounted for by the assumption that the male sex stimulates the instability. Experiments are now in progress which are expected to throw more light on this problem.

Summary.—Miniature-3 gamma gene is unstable in somatic cells. The results of experiments in which the flies carrying this gene were reared at 30 ± 0.2, 25 ± 1 and 20 ± 0.2 degrees centigrade, respectively, are interpreted to indicate that these temperature differences have not produced any effect on the instability of the gene.

It has been found that the frequency of gene changes was alike in males and in females in spite of the fact that a female carries approximately twice the number of miniature genes carried by a male.

[1] Muller, H. J., *Genetics*, **13**, 279–357 (1928).
[2] Goldschmidt, R., *Biolog. Zentralblatt*, **49**, 437–448 (1929).
[3] Jollos, V., *Ibid.*, **50**, 541–554 (1930).
[4] Demerec, M., *Jour. Gen.*, **24**, 179–193 (1931).
[5] Andersson-Kottö, I., *Zeitschr. ind. Abst. Vererbungslere*, **56**, 115–201 (1930).
[6] Demerec, M., these PROCEEDINGS, **15**, 834–838 (1929).

AN INTERCHANGE IN MAIZE GIVING LOW STERILITY AND CHAIN CONFIGURATIONS

BY C. R. BURNHAM[1]

WILLIAM G. KERCHHOFF LABORATORIES OF THE BIOLOGICAL SCIENCES, CALIFORNIA INSTITUTE OF TECHNOLOGY

Communicated April 26, 1932

The culture of maize which gave semisterile-1 plants (Brink, 1927) contained also two plants with a low percentage of pollen abortion (10–18 per cent based on counts made at that time). This culture had not been x-rayed. Low steriles appeared in the first generation crosses obtained from one of these plants. Pollen counts show them to be about 25 per cent sterile. Thirty-seven plants were counted, giving a total of 58,000 pollen grains, of which 26.3 per cent were practically devoid of starch.

Root tip counts showed that the low sterile plants have 20 chromosomes, demonstrating that this low sterile line differs from the 21 chromosome low steriles arising from irregular (3–1) disjunction of the chromosomes in the rings of semisteriles (Burnham, 1930, McClintock, 1931).

Diakinesis configurations in the microsporocytes of low sterile plants showed 8 bivalents plus a chain of four chromosomes, while their normal sibs had 10 bivalents. In the low sterile plants, a chain of four chromo-

somes is formed regularly. No case has been observed of a ring of four, nor any breaking of the chain to form "pairs."

The chain is associated with the nucleolus. This indicates that the satellite chromosome is involved, since in *Zea mays*, this chromosome is the only one which is attached to the nucleolus.

In order to identify the other chromosome involved in the chain, the low sterile was crossed with homozygous interchange types in which the chromosomes involved had been determined. The low sterile was used as the pollen parent to avoid any complication from $n + 1$ gametes which sometimes are formed from a 3–1 disjunction of four associated chromosomes. It has been found from a study of trisomic plants that $n + 1$ pollen functions only rarely in competition with n pollen (McClintock and Hill, 1931). Crosses of the low sterile with semisterile-1 (an interchange between the *P-br* and the *B-lg* chromosomes, Brink and Cooper, 1931) gives a chain of 6 chromosomes plus 7 bivalents, indicating that one of these two chromosomes is involved. The cross with semisterile-3 (an interchange between the *P-br* and the gl_1-*ra* chromosomes, unpublished data of the author) also gives a chain of 6 chromosomes, indicating that one of these two is concerned. Since the *P-br* chromosome is the one common to semisterile-1 and semisterile-3, it must be the one involved in the low sterile.

The chain of 4 in the low sterile therefore involves the satellite chromosome, carrying the genes of the *Y-pl* linkage group (McClintock, 1932); and the *P-br* group which belongs to the longest chromosome (Burnham, 1930).

In order to obtain evidence regarding the nature of the chromosomal change, pachytene stages in microsporocytes of the low sterile plants were studied. McClintock, 1930, and Brink and Cooper, 1931, have shown that the chromosomes at this stage are very much longer than at diakinesis and are closely synapsed. The interchanges give a cross-shaped synaptic complex resulting from synapsis of homologous parts.

The pachytene configuration in the low sterile plants is definitely not a cross-shaped complex but approximates a T shape.

The normal satellite chromosome is represented in figure 1*a*. At the pachytene stage, according to McClintock's (1931 and unpublished) description, it consists of a long arm beyond the spindle fibre insertion region, a short section from the insertion region to an enlarged reticulate region, followed by a very lightly stained, almost colorless region, then the satellite proper. The "colorless" and reticulate regions are attached to the nucleolus. The satellite proper is composed of 4 chromomeres, i.e., a basal chromomere adjacent to the "colorless" region, two small intermediate chromomeres, and a much larger distinctive terminal chromomere (see *a*, Fig. 1). In many figures the two intermediate chromomeres are so close together that they appear as one.

The *P-br* chromosome has almost a median insertion region (Fig. 1*b*). The relative length of the two arms is 1:1.2+ (McClintock, 1929, and from prophase measurements in these studies). In the pachytene stage, the arms can be distinguished readily, since on the short arm the region adjacent to the insertion appears to be more darkly stained.

In the low sterile plants, the pachytene figures clearly indicate that an

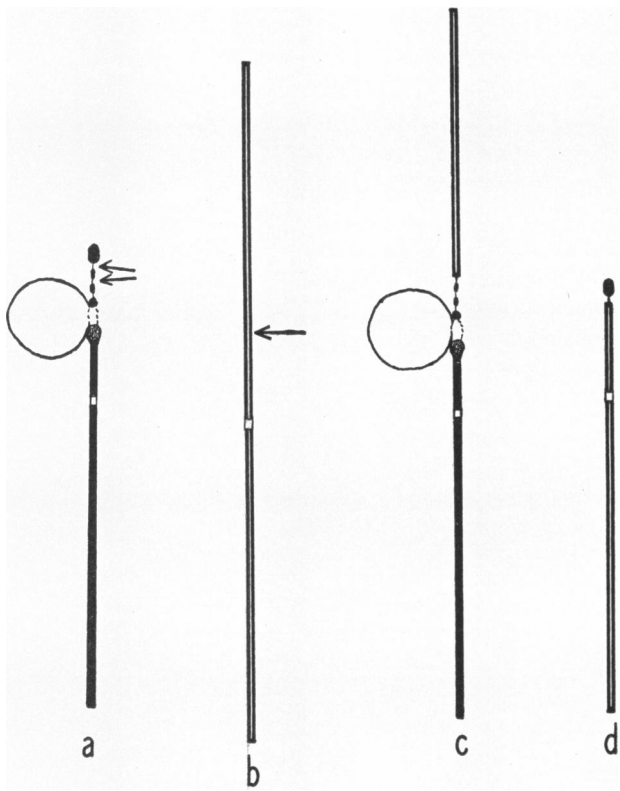

FIGURE 1

a—normal satellite chromosome; *b*—normal *P-br* chromosome; *c*, *d*—the new chromosomes resulting from an interchange at the positions of the arrows in *a* and *b*. The end of the satellite has interchanged with a piece of the longer arm of *P-br*.

interchange has occurred at the positions indicated by the arrows in figure 1, *a* and *b*. Several figures were obtained in which these chromosomes could be followed for their entire lengths. Many more were found which were clear for the interchange region. The break in the *P-br* chromosome is about $^3/_4$ of the distance from the end of the longer arm, that in the satellite chromosome is somewhere between the second and end chromomeres of

the satellite itself. In all probability, only the terminal chromomere is involved in the interchange. It is certain that it includes no more than two chromomeres of the satellite. The two chromosomes resulting from the interchange are represented in figure 1, *c* and *d*. The synaptic configuration observed in plants heterozygous for the interchange is given in figure 2. The distinctive end chromomere on the normal satellite chromosome and its homolog on the interchange chromosome are represented as unsynapsed. Although this chromomere can be readily identified, no case of synapsis of these two homologous chromomeres was observed. Thus, although an interchange had occurred, the configuration at pachytene is practically a T.

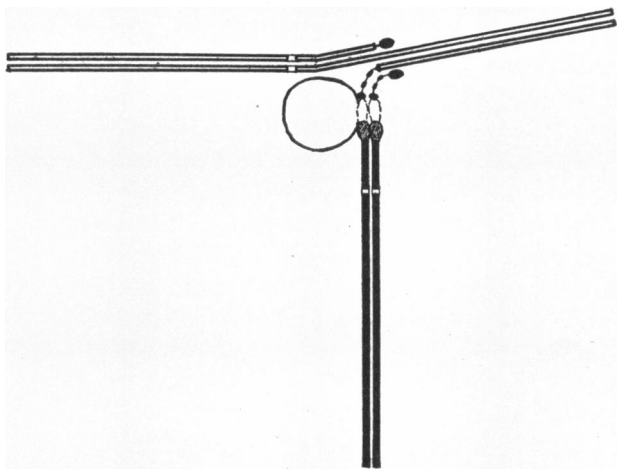

FIGURE 2

Diagram showing the pachytene configuration in a plant heterozygous for the interchange. The terminal knobs of the satellite fail to synapse.

The chain at diakinesis is presumably the result of this earlier lack of synapsis.

In the interchanges which involve large segments of chromosomes, the sterility approximates 50 per cent. The interchange here described shows but 25 per cent pollen sterility. The low sterility might be due to a distribution of the four chromosomes at anaphase I which decreased the proportion of abortive types. It should be possible to determine the type of distribution from the configurations at metaphase I. As shown above, synapsis to form a ring of 4 probably always fails at one point, namely the terminal chromomere of the satellite. Therefore the order of the chromosomes in the chain should be constant. This makes it unnecessary to recognize the four chromosomes to determine the type of distribution. If they

are numbered 1,2,3,4 starting at one end of the chain, 2-by-2 distribution might be 1 4-2 3 (adjacent), 1 3-2 4 (alternate) or 1 2-3 4 (adjacent). None of the latter type of adjacent distribution was observed. Of 379 metaphase I figures in which the type could be determined, 180 were alternate, 199 were adjacent. The alternate distribution gives rise to two nuclei, each possessing the full chromatin complement and should be fully fertile. The adjacent distribution gives rise to two nuclei, both of which are deficient, one for a long segment of the *P-br* chromosome, and the other for one or two chromomeres of the satellite. Since the alternate and adjacent distributions occur with about equal frequency, the sterility should be about 50 per cent if both types of deficiencies result in abortive spores. The type which is deficient for a part of the satellite might not abort, since it involves such a minute part of the monoploid complement. In that case, only 25 per cent sterility would be expected. Since this deficient type also carries a duplication for a large section of the longer arm of the *P-br* chromosome, it would be expected to function rarely, if at all, through the pollen (see page 1). Therefore in determining if this type of gamete functions, progeny of crosses with normal plants using the low sterile as the female parent were studied. From the observations on anaphase I distributions given above, the plants should be of three types: One showing 10 bivalents at diakinesis and giving normal pollen, one showing a chain similar to that of the female parent and giving about 25 per cent sterility, and one which is monosomic for the terminal one or two chromomeres of the satellite and trisomic for a long segment of the longer arm of the *P-br* chromosome. These three types should occur with equal frequency. The observed results were 9:19:1, respectively. The one plant which resulted from the functioning of a gamete with the small deficiency was readily distinguishable at diakinesis. Contrasted with the ordinary low sterile type in which chains are always formed, this plant forms chains and "pairs." "Pairs" were formed somewhat more frequently than chains, actual counts giving 213 "pairs" : 151 chains. The satellite "pair" is unequal, one member being much longer than the other due to the addition of the long piece of the *P-br* chromosome in place of the small terminal portion of the satellite. The other nine pairs are normal.

The pollen on this one plant having the small deficiency showed about 2 to 3 per cent sterility, the amount usually found in normal plants. It also appears to have a normal set of seed. This clearly demonstrates that the 25% sterility in the type which always forms chains is a result of non-abortion of the microspores deficient for the small piece of the satellite. It also shows that this type of gamete may survive and function in the ovules. The reason for the deviation from the expected 1:1:1 ratio is not clear. A possible explanation may be differential viability. Germination in this culture was only 66 per cent. Since this new type is trisomic for a part

of the *P-br* chromosome, it may be weaker than its sibs and be eliminated in a culture showing poor germination. The test is being repeated.

In the progeny from selfing a low sterile plant, the type homozygous for the interchange has been isolated. At diakinesis in this type there are ten pairs. The interchange chromosome attached to the nucleolus now has a long arm on either side of its point of attachment. The resulting chromosome is much longer than any of the other chromosomes. In prophases, the other interchange chromosome clearly shows that it is homozygous for the distinctive terminal knob of the satellite on the end of what is now the short arm.

Data on the inheritance of low sterility are too meager to draw any definite conclusion. The crosses with standard normal lines using the original low sterile plant as the pollen parent gave only a small proportion of plants showing low sterility. One plant from the second generation gave similar results in the cross of normal × low sterile, the total being 260 normal : 40 low sterile. In the third generation, several plants gave very close to a 1:1 ratio. In cultures studied cytologically, one gave 12 plants with 10 bivalents and 21 plants showing a chain of 4; another gave 10 : 6. In all cases, only the plants having chains showed sterility in the pollen. The original pollen classification was rechecked independently. No errors were found. A tentative explanation of the deviations from the theoretical 1 : 1 ratio is that a factor similar to that causing low sugary ratios (Emerson, 1925, Mangelsdorf and Jones, 1926) may have been present at some distance from the break in either of the two chromosomes. Crossovers would make it possible to isolate lines giving 1 : 1 ratios as well as those giving a high proportion of low steriles.

The fact that the gamete carrying the small deficiency may function furnishes a means of testing genes for their presence in this portion of the chromosome. The one plant which arose from a gamete of this type was from the cross of low sterile × albescent. According to cytological and genetical evidence reported by McClintock (1932), the order must be satellite-albescent-*Y-Pl*, *Y* and *Pl* being definitely located on the long arm. The plant obtained was not albescent, indicating that this factor is not located in the terminal chromomere of the satellite.

If this same deficient gamete which also carries a duplication for a part of the *P-br* chromosome does function in its theoretical frequency where germination is good, selfs of low sterile plants heterozygous for factors in the *P-br* chromosome should give trisomic ratios for those factors which lie in the arm involved in the interchange and disomic ratios for the remainder.

The writer's cytological observations on semisterile -3 in which the interchange also involves the longer arm of *P-br;* together with the genetic linkage observed there with brachytic indicate that the low sterile reported here will also show close linkage with brachytic. The tests are in progress.

Discussion.—In the semisteriles where critical cytological evidence has been obtained, there has been an interchange involving relatively large sections of the chromosomes (McClintock, 1930, Brink and Cooper, 1931, Rhoades, 1931). In these cases, a ring of four chromosomes is formed at diakinesis. Disjunction of the four chromosomes at anaphase is such that approximately 50 per cent of the spores are deficient for relatively large sections of the monoploid complement; thus accounting for the sterility in these types. The type heterozygous for the attachment of a fragment of one chromosome to a non-homolog, termed "simple translocation" in Drosophila, would be expected to give a T-shaped configuration at pachytene, a chain in place of a ring at diakinesis (see note at end), and only 25 per cent sterility. Brink and Cooper (1931) in an abstract have described a case assumed to be of this type. The observations presented in this paper demonstrate that a type which is definitely an interchange may exhibit all these features.

It is probable that some of the types suspected of being simple translocations in Drosophila would prove to be interchanges if critical cytological evidence could be obtained.

In the case reported here where one of the interchanged pieces is short, chain configurations are formed at diakinesis. If both pieces were very short, ten "pairs" might be formed regularly, giving no evidence that an interchange had occurred.

Note.—The author has pointed out (Burnham, 1930) that a "simple translocation" might give rise to rings in place of chains as a result of 4-strand crossing-over. This would occur only if the opening out at diakinesis were equational at the spindle fibre region. The case described here in which chains are always formed at diakinesis may be taken as evidence that the opening out is reductional at the spindle fibre region.

[1] NATIONAL RESEARCH COUNCIL FELLOW, California Institute of Technology, Pasadena.

REFERENCES

Brink, R. A., *J. Hered.*, **18**, 266–270 (1927).
Brink, R. A., and D. C. Cooper, *Genetics*, **16**, 595–628 (1931).
Brink, R. A., and D. C. Cooper, Abstract in *Anatom. Record*, **51**, 107–108 (1931).
Burnham, C. R., *Proc. Nat. Acad. Sci.*, **16**, 269–277 (1930).
Emerson, R. A., Abstract in *Anatom. Record*, **29**, 136 (1925).
Mangelsdorf, P. C., and D. F. Jones, *Genetics*, **11**, 423–455 (1926).
McClintock, B., *Science*, **69**, 629 (1929).
McClintock, B., *Proc. Nat. Acad. Sci.*, **16**, 791–796 (1930).
McClintock, B., *Proc. Nat. Acad. Sci.*, **17**, 485–491 (1931).
McClintock, B., and H. E. Hill, *Genetics*, **16**, 175–190 (1931).
McClintock, B. Missouri Agr. Exp. Sta. Res. Bull. 163 (1931).
Rhoades, M. M., *Proc. Nat. Acad. Sci.*, **17**, 694–698 (1931).